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Environmental and Biological Effects Following Application of Diazinon Impregnated Strips Within a Laboratory Animal Room

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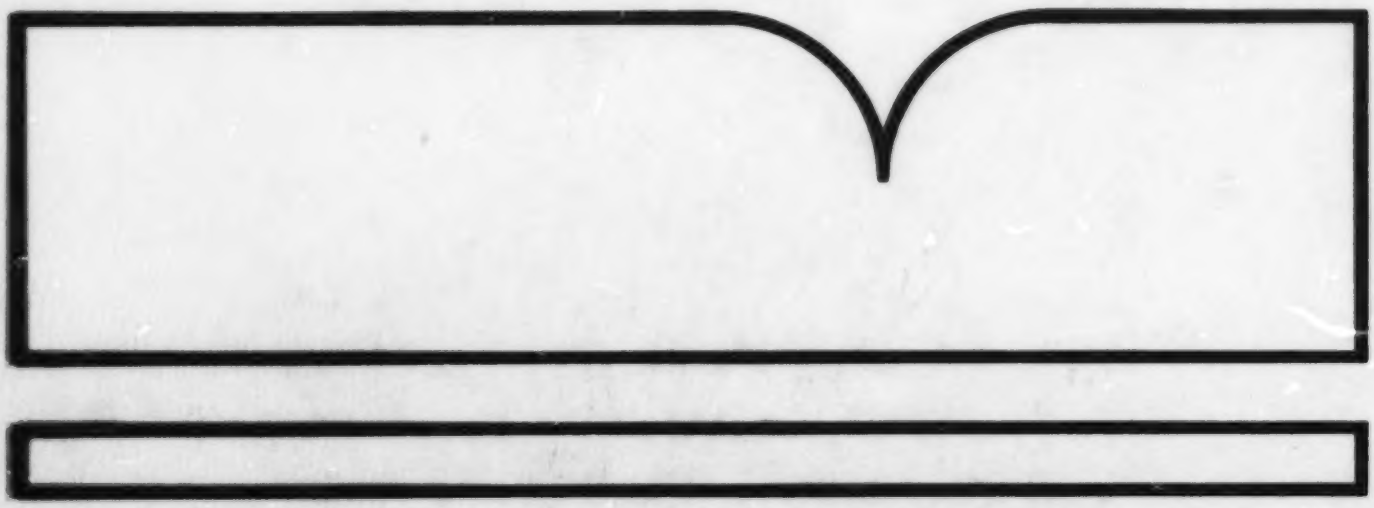

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16. ABSTRACT

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Environmental and Biological Effects Following Application of Diazinon Impregnated Strips Within a Laboratory Animal Room[1,2,3]

Donald K Hinkle, Joseph E Suggs, and Merrill D Jackson

Summary | Rats were exposed to diazinon impregnated strips in a conventional laboratory animal room. The air in the room was monitored for the pesticide. Erythrocyte acetylcholinesterase and plasma pseudocholinesterase activities were determined periodically. Air concentration of the pesticide never exceeded 1.34 μg/m³. No significant change in enzyme activites were observed.

Key Words | Diazinon — Air monitoring — Cholinesterases — Rat

Increasing concern for improving the quality of the environment has led to a marked reduction in the use of the more persistent insecticides, such as chlorinated hydrocarbon compounds. They largely have been replaced by the organophosphate and carbamate compounds. These two classes of chemicals are highly toxic in their concentrated form and, therefore, may pose a significant health hazard to commercial pest control operators. To reduce this hazard, some pesticide formulators have introduced new application technologies which largely eliminate the requirement for handling of highly concentrated chemicals. One such product is a multilayered polymeric plastic strip measuring 2.5 x 10 cm and impregnated with a 10% concentration of the insecticide diazinon (0,0-diethyl 0-(2-isopropyl-6-methyl-4-pyrimidiryl) phosphorothioate).[4]

This product is currently registered by the US Environmental Protection Agency for use by commercial pest control operators in control of crawling insect pests by contact. It is, therefore, possible that it may find its way into research animal quarters as part of a commercial pest control program. This study was undertaken to determine ambient air concentrations and any erythrocyte cholinesterase or plasma pseudocholinesterase alterations in laboratory rats as a result of treating a conventional animal room with these pest control strips to control crawling insects.

Materials and Methods

Laboratory animal room: Eighty 90-day-old Sprague-Dawley rats [Crl:COBS CD (SD)],[5] 40 males and 40 females, were housed singly in stainless steel wire mesh suspended rat cages, five tiers high and distributed equally within four cage holding racks. The animal holding room measured 3 x 3.6 m with a 2.4 m ceiling height. There were an estimated 8-10 air changes per hour with a single air inlet and a single air exit (approximately 3 m apart) located at ceiling level. The rats were kept in an environment maintained at 21 ± 1.0°C with a relative humidity of 50 ± 20%.

Adhesive backed, polymeric plastic strips containing a 10% concentration of diazinon and measuring 2.5 x 10 cm were attached to the perimeter of the room at approximately 0.75 m intervals. Eighteen strips were placed on the wall at floor level, and another 18 strips were placed 1 m up on the wall.

Air concentrations of diazinon were collected during two experiments and were

[1]From the Environmental Biology and Environmental Toxicology Divisions, Health Effects Research Laboratory, US Environmental Protection Agency, Research Triangle Park, NC 27711.
[2]Use of trade names is for identification only and does not constitute endorsement by the US Environmental Protection Agency.
[3]Reprint requests to: Donald K Hinkle, Mannheimer Foundation, 20255 S W 360 Street, Homestead, FL 33030.
[4]Hercon Insectape, Health-Chem Corp, New York, NY.
[5]Charles River Breeding Labs, Wilmington, MA.

averaged. Enzyme activity levels were measured only during the second experiment.

Air sampling: Air samples were collected with a portable, high-volume sampler.[6] The sampler was positioned on the floor in the center of the room at a distance of 1.0 m from the animal cages. The air intake of the air sampler was positioned 28 cm from the floor surface. One side of the dual-intake sampler was blocked so that the room air was sampled at 11.2 m^3/hour through a single vapor trap. The diazinon vapor trap consisted of a 15-cm circular composite filter pad (1,2) composed of a 3:1 mixture of porous polymer[7] and glass fibers sandwiched between two glass fiber filters. This trap has been employed previously in the collection of organophosphorus pecticides (1,3).

Sampling periods were for 2 hours each at 0.25, 1, 2, 3, 4, 7, 15, and 30 days after installation of the pest control strips. Background samples were taken prior to strip installation. After each collection, the filter pad was stored at −17°C in a sealed container until it was analyzed. This storage period was less than 15 days.

Analysis of air samples: All solvents used were glass distilled. The standard diazinon was of 89% purity.[8]

Each filter pad was individually extracted with 800 ml of ethyl acetate in a Soxhlet extractor for 4 hours at five cycles per hour. The extract was concentrated to approximately 1 ml in a rotary evaporator and quantitatively transferred to a 15 ml centriguge tube (graduated and glass stoppered). Dilutions as needed for gas chromatography were made with ethyl acetate.

Gas chromatography was performed on a 6.4-mm x 183-cm glass column. The column packing was 4% SE-30 and 6% OV-210 (liquid phase) on white diatomaceous earth[9] mesh ranges of 80 to 100 (solid phase). The gas chromatograph was operated at the following temperatures: column oven, 200°C; injection port, 220°C; and detector oven, 180°C. A nitrogen flow of 100 ml/minute was maintained through the column and detector. A flame photometric detector was operated in the phosphorus mode. Air at a flow rate of 80 ml/minute and hydrogen at a flow of 60 ml/minute were supplied to the detector.

Determination of sampling and analysis efficiencies: Collection efficiency was measured by introducing diazinon vapors from a fortified 15-cm circular glass fiber prefilter adapted to the intake 1 cm in front of the vapor trap. The prefilter was fortified by dropwise addition of 2–40 μg of diazinon in benzene, allowed to dry for 15 minutes, then placed on the sampler. After 2 hours of operation at 11.2 m^3/hour, the prefilter and vapor trap were separately analyzed for diazinon. A collection efficiency of 93% ± 6% was calculated from four determinations. Analytical recovery from fortified filter pads was 95% in two determinations. The overall sensitivity of the method was estimated to be better than 0.01 μg/m^3.

Analyses of blood samples: Erythrocyte acetylcholinesterase and plasma pseudocholinesterase activities were determined by an automated pH stat method (4). The erythrocytes and plasma were separated by centrifugation. The erythrocytes were washed three times with 0.9% saline, and a 10% hemolysate was prepared with distilled water. The hemolysate (0.2 ml) was added to 4.7 ml of 0.15 N saline, and the pH was adjusted to 8.0 in the reaction vessel. Acetylcholine was used as the enzyme substrate, and activity was determined by tritration of the released acetic acid. Plasma pseudocholinesterase activity was determined in a similar manner by the addition of 0.1 ml plasma to the reaction vessel. A recording pH stat was used employing 0.01 N sodium hydroxide as the titrant.

Whole blood samples were collected via intracardiac puncture from five individual rats of each sex on days 1, 3, 7, 15, and 30. Heparin was used as an anticoagulant. Control erythrocyte acetylcholinesterase and plasma pseudocholinesterase activities were obtained from unexposed rats.

Results

The detectable residues of diazinon recovered from filter media used in ambient air sampling procedures are presented in Table 1. All residue levels were at concentrations of less than 1.34 μg/m^3 of air. Peak ambient air concentration was recorded at the Day 15 sampling period. The levels of environmental contamination by diazinon reported here were below human olfactory sensitivity. Conversely,

[6]Environmental Research Corporation, St Paul, MN.
[7]Porapak®, Waters Associates, Milford, MA.
[8]Pesticide Repository, US Environmental Protection Agency, Research Triangle Park, NC.
[9]Gas Chrom Q. Applied Science, State College, PA.

Table 1

Diazinon in air of laboratory animal room treated with diazinon impregnated strips

Time after treatment (days)	Corrected air concentration ($\mu g/m^3$)
0[a]	0.01
0.25	0.32[b]
1	0.56
2	0.76
3	0.60
4	0.84
7	0.90
15	1.34
30	1.21

[a]Background in room before treatment
[b]Average of two separate experiments

more traditional insecticide application techniques usually result in significant olfactory stimulation. The olfactory stimulation is due to the hydrocarbon carrier normally used; however, with the pesticide strips there was no hydrocarbon carrier present.

Acetylcholinesterase activities in the rat erythrocytes are reported in Table 2.

Table 2

Erythrocyte cholinesterase activity of rats exposed to airborne diazinon

Time after treatment (days)	Cholinesterase activity (μmoles/min/ml)[a,b] Male	Female
Pretreatment control	1.37 ± 0.25	1.33 ± 0.09
1	1.34 ± 0.28	1.53 ± 0.12
3	1.62 ± 0.08	1.62 ± 0.16
7	1.59 ± 0.15	1.51 ± 0.22
15	1.54 ± 0.22	1.48 ± 0.18
30	1.41 ± 0.34	1.50 ± 0.22

[a]All values are the average of five rats (mean ± standard deviation)
[b]μmoles of acetate released

Table 3

Plasma pseudocholinesterase activity of rats exposed to airborne diazinon

Time after treatment (days)	Cholinesterase activity (μmoles/min/ml)[a,b] Male	Female
Pretreatment control	0.43 ± 0.06	0.41 ± 0.06
1	0.42 ± 0.06	0.42 ± 0.07
3	0.46 ± 0.05	0.39 ± 0.06
7	0.40 ± 0.06	0.39 ± 0.03
15	0.40 ± 0.07	0.43 ± 0.08
30	0.41 ± 0.05	0.44 ± 0.06

[a]All values are the average of five rats (mean ± standard deviation)
[b]μmoles of acetate released

Table 3 contains the plasma pseudocholinesterase activities. The plasma pseudocholinesterase and erythrocyte acetylcholinesterase activity in both sexes was not inhibited.

Discussion

The experimental design was intended to: (1) determine the ambient air concentration in a laboratory animal room treated with a measured quantity of diazinon contained in a contact delivery system, and (2) to determine if an animal room treated with these pest strips would result in detectable alteration in erythrocyte or plasma cholinesterase activity.

Based upon the results of our sampling of ambient air through the 30-day period of this experiment, it appears that the maximum concentration of diazinon occurs between Day 15 and Day 30 following application, (Table 1). The pesticide formulation used in this study contained a contact poison designed to kill crawling insect pests only, and it was contained in a polymeric plastic delivery system designed to minimize volatalization of the active ingredient. The usual commercial application procedures for contact pesticides is by means of an aqueous carrier and will likely result in significantly higher ambient air concentrations than were reported herein.

Our data indicates that neither plasma pseudocholinesterase nor the erythrocyte acetylcholinesterase activity of the rat was affected by the levels of diazinon released by the diazinon-impregnated strips. However, the decision to apply any chemical pest control within laboratory animal rooms should have the prior approval of both the facility director as well as the scientists involved.

References

1. Lissick M O, Bosin W A: *Development of an Efficient High-volume Sampling System for Alkyl Phosphonate Vapors at the ppb Concentration level.* Report No 8-7613. Environmental Research Corporation, St Paul, 1975

2. Lewis R G: Sampling and analysis of airborne pesticides. In *Air Pollution from Pesticides and Agricultural Processes*, p 52–94, Lee R E, Ed, CRC Press, Cleveland, 1976

3. Jackson M D, Lewis R G: Volatization of methyl parathion from fields treated with microencapsulated and emulsifiable concentrate formulations. *Bull Environ Contam Toxicol* 21:202–205, 1979

4. Nabb D P, Whitfield F: Determination of cholinesterase by an automated pH stat method. *Arch Environ Health* 15:147–154, 1967